 ISSN 1473-2262

The notochordal cell in the postnatal intervertebral disc

C. J. Hunter[1]

[1] *University of Calgary, Calgary, Alberta, Canada*

INTRODUCTION: During embryogenesis of the intervertebral disc, the cells of the notochord play a critical role in initiating tissue formation, and may be directly responsible for development of the nucleus pulposus. In some species, including humans, these notochordal cells may eventually be lost, perhaps through apoptosis or terminal differentiation, and are replaced by chondrocyte-like cells[1]. However, there is some evidence that the notochordal cells may persist in at least some humans.

The nucleus pulposus of the intervertebral disc undergoes substantial changes during aging and degeneration, which can compromise disc function and lead to chronic pain and debility. One of the first morphological changes is the loss of the notochord-derived cells[2]. However, the significance of loss of the notochordal cells and subsequent degeneration of the disc has not been thoroughly studied.

METHODS: We have used numerous tools, including confocal scanning microscopy, transmission electron microscopy, cryogenics, immunohistochemistry, and real-time PCR to investigate the functional behavior of adult-derived notochordal cells.

RESULTS: Both embryonic notochord and adult notochordal cells have been long described as "physaliferous," meaning they appear "foamy" in conventional histological sections[3]. We have found that while this term is accurate, it only begins to describe the structure of these unique cells. The cells contain massive vacuoles which take up as much as 80% of the cell volume (Fig. 1), surrounded by dense actin cortices and multilamellated membranes (Fig. 2)[4]. These vacuoles appear to function at least in part in osmoregulating the cells[5]. The cells are occasionally multinucleated, a phenomenon which can become more common *in vitro*. *In situ*, the cells exist in tightly joined clusters of up to 100 cells (Fig. 3). Within these clusters, the cells are connected via gap junctions, and maintenance of these connections seems to be important for *in vitro* survival[5].

In situ, the notochordal cells do not exist in isolation. We have begun to identify interactions between notochordal cells and other cell types, including chondrocytes, annulus fibrosus cells, and mesenchymal stem cells. Notochordal cells stimulate gene expression in these cell types, particularly genes for small proteoglycans.

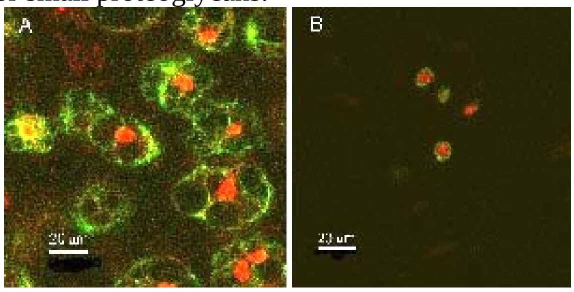

Figure 1. Actin structure of disc cells, (A) young disc (B) aged disc. Note the presence of large vacuoles in the young cells. Green: actin; red: cell nuclei. Bar: 20 µm.

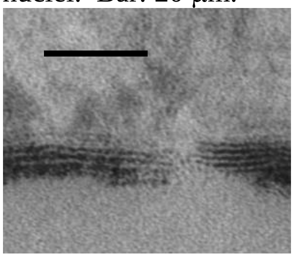

Figure 2. TEM of the multilamellated membrane around notochordal cell vacuoles. Bar: 500 nm.

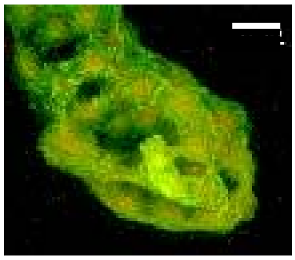

Figure 3. 3D reconstruction of a notochordal cell cluster containing over 30 cells. Bar: 20 µm.

DISCUSSION & CONCLUSIONS: While we have only begun to scratch the surface, the results thus far are quite intriguing. The fundamental questions which face us as a research community are (1) what function do the notochordal cells serve after formation of the axial skeleton? (2) why do the notochordal cells disappear during maturation? (3) do the notochordal cells and their gelatinous matrix serve a role in the pathogenesis of intervertebral disc degeneration?

REFERENCES: (1) Butler, in *The Biology of the Intervertebral Disc*, 1989. (2) Urban *et al*, *Am Zool*, 2000. (3) Trout *et al*, Tiss Cell, 1982. (4) Hunter *et al*, J Anat 2003. (5) Hunter *et al*, Trans ORS 2005. (6) Hunter *et al*, Spine 2004.

ACKNOWLEDGEMENTS: This work was supported by the Canadian Institutes of Health Research under grant number 64385.